Analysis of retinal light adaptation with the flicker electroretinogram

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To study retinal light adaptation we measured and analyzed the flicker electroretinogram response to stimuli that varied in temporal frequency, retinal illuminance, and modulation depth. The responses measured at 100% modulation showed the classic adaptation pattern, being independent of mean retinal illuminance at low temporal frequencies, consistent with Weber adaptation, and increasing in proportion to mean retinal illuminance at high temporal frequencies, consistent with linearity. At 25% modulation, however, high-frequency linearity was not found. The response amplitude consistently showed a minimum at 40–48 Hz. When modulation was systematically varied, response amplitudes measured at 16 and 22 Hz showed Weber adaptation at all modulations and response phase was relatively constant with modulation, whereas response amplitudes at 40 and 48 Hz showed adaptation at low modulations but linearity at high modulations and response phase varied with modulation. We conclude that retinal gain controls also operate at high temporal frequencies.

Key words: light adaptation, linearity, temporal sensitivity, flicker electroretinogram, retinal gain control.

1. INTRODUCTION

Light adaptation involves changes both in the sensitivity and in the temporal dynamics of the visual system. It is generally believed that with increasing illuminance the eye becomes relatively less sensitive to mean light level but more sensitive to fast flicker.[1–3] This relation was formulated by Kelly[2] as a transition from Weber adaptation to linearity with increasing temporal frequency. That is, human visual sensitivity is inversely proportional to the mean light level at low temporal frequencies (i.e., $\Delta I/I$ = constant, Weber's law) but is determined mostly by the absolute amplitude of modulation (ΔI) at high temporal frequencies (linearity). Although Kelly's original analysis was based on psychophysical measurements of flicker thresholds, the same pattern of results was also obtained from electrophysiological studies of retinal responses, such as in monkey ganglion cells (e.g., Purpura *et al.*[4]) and in the human focal electroretinogram (ERG; e.g., Seiple *et al.*[5]). From the electrophysiological results it has been suggested that the differential effects of light adaptation on the temporal response must occur at the retinal level and, in the case of the study of Seiple *et al.*, at the level of photoreceptors.

In Kelly's frequency analysis the main features of temporal adaptation are characterized by a trade-off between sensitivity and temporal dynamics. This is a theoretically important formulation. Many visual gain control models derived from physiological measurements are based on these concepts and employ either feed-forward or feedback circuits to produce this type of behavior.[4,6–8] To achieve this pattern of results these models incorporate gain controls that do not operate at high temporal frequencies. Single or sequential filters can implement such frequency-dependent effects.

Kelly's analysis of the flicker response was later expanded by Tyler,[9] who reexamined the flicker threshold measurements by using a luminance analysis approach. Tyler concluded that there is a trade-off in the contribution of different cell populations with changes in retinal illuminance. These cell populations are presumably the sustained and transient retinal ganglion cells, or the P and M cells as they were later classified. The sustained or P cells are sensitive to flickering edges, whereas the transient or M cells are sensitive to changes in mean luminance. These cells have different temporal responses as well as different luminance responses. With small-field flicker[1] the sustained or P cells were hypothesized to be responsible for the low-luminance mechanism and the transient or M cells were responsible for the high-luminance mechanism at high temporal frequencies. With large-field flicker[2] a single luminance function was sufficient to fit all the data, which corresponded to the high-luminance, transient mechanism. Thus Tyler's luminance analysis involves a two-mechanism model at least for the small-field data, with the properties of both mechanisms matching the known physiology.

Although both Kelly's and Tyler's analyses of the psychophysically determined flicker thresholds yielded important understanding of temporal adaptation, it is not known whether these analyses can be applied to retinal responses when stimulus modulation is systematically varied. In the present study we examine frequency-dependent adaptation by using the flicker ERG. Our approach is different from that of Seiple *et al.*[5] in that, instead of using a response extrapolation procedure for determining a threshold, we measure the flicker ERG responses at a series of modulations. We adopt this approach because the response amplitude of the flicker ERG depends on modulation in a highly nonlinear, complex fashion[10–12]; thus the results obtained by a linear extrapolation to a response amplitude of zero represent a special case.

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There are three advantages in using the flicker ERG. First, the flicker ERG is a measure of the response of the photoreceptors and bipolar cells, as it has been established that the flicker ERG shows functional properties of the photoreceptors,[5,10,13–15] has peak current source density in the distal retina,[16] can be directly compared with the local receptor potentials,[17,18] may contain a contribution from bipolar cell responses,[19,20] but does not reflect responses from nonneural elements such as glial cells because of their slow response.[21] Second, studies that use a linear systems analysis approach have found multiple frequency components in the flicker ERG.[11,22] One of these components operates at frequencies below 10 Hz and reflects primarily responses of the rod system. The other two, with peak sensitivities at 20 and 40 Hz, respectively, reflect primarily responses of cone systems and have different response properties with changes in stimulus parameters. These results indicate that different frequency regions can be distinguished in the flicker ERG. Finally, measurement of the flicker ERG allows us to characterize the temporal responses of the retina over a range of modulation depths that are unavailable when psychophysical threshold measurements are used. In a previous study[12] we showed a strong interaction between temporal frequency and stimulus modulation in the flicker ERG. The fundamental response amplitude of the flicker ERG showed a compressive relation to modulation at 16 Hz, an accelerating relation at 40 and 48 Hz, and a linear relation at 64 and 72 Hz. The result indicated that there is a modulation-dependent nonlinear adaptation mechanism in the retina. In the present study we further investigated how stimulus modulation affects light adaptation. To do this we measured the fundamental response amplitudes of the flicker ERG to stimuli that varied in retinal illuminance, temporal frequency, and modulation depth and tested the results against the predictions from Weber adaptation and linearity, using an analysis similar to Kelly's.[2] Although the results reported below are based on an analysis of the fundamental response component to sine wave flicker, it is important to note that in general, for a nonlinear system, both linear and nonlinear units contribute to the fundamental response.

2. METHODS

A. Subjects

The two authors served as subjects. Both had normal vision with no evidence of ocular disease. The experimental protocols were reviewed and approved by the local institutional review board.

B. Apparatus and Stimuli

The apparatus used in this study was described previously.[23] Briefly, it consists of a two-channel Maxwellian-view system that uses red (633-nm) and green (543-nm) He–Ne lasers as light sources. The mixture of the red and the green lights was close to unique yellow, and the two lights were modulated in phase throughout the experiment so that the subjects saw a uniform, flickering yellow field. The maximum retinal illuminance for both the red and the green lights was 3.8 log Td. Between conditions, the mean retinal illuminance of the stimulus was varied by a calibrated neutral-density wedge. Within a condition, the time-averaged retinal illuminances of both the red and the green lights were constant during modulation. The modulation depth and the temporal frequency of the yellow field were controlled by a programmable function generator. The field size was 40 deg in diameter. The 40-deg field was viewed inside a white ganzfeld produced by a Goldmann–Weekers adaptometer to suppress stray light responses. The luminance of the ganzfeld surround was adjusted for each condition to match approximately the luminance of the stimulus field. The output of the stimulus, calibrated by a photodetector placed at the exit pupil of the optical system, was linear to the resolution of the recording system.

C. Signal Acquisition and Analysis

ERG responses were recorded with DTL thread electrode. Responses were filtered and amplified by two sequential amplifiers (Grass P511K), and both were set to pass 3–300 Hz. The gain for the first amplifier was 1000, and for the second, 200. The output of the second amplifier was digitized by a 12-bit analog-to-digital converter (Data Translation) at a sampling rate of 512 Hz.

A total of 2048 samples were recorded for each response (4-s epoch). Depending on the signal amplitude, either 8 or 16 responses were time averaged for each trial. Both the sampling and the averaging were synchronized with the stimulus through a single master clock. The averaged responses were analyzed off line with a discrete Fourier transform. The magnitude, the phase, and the signal-to-noise (S/N) ratio were computed at the stimulus frequency and its second and third harmonics. We estimated the S/N ratio by computing the ratio of the response at the frequency of interest to the averaged response at nearby frequencies (i.e., signal-plus-noise to noise ratio). The noise was estimated as the average power in the 10 frequency bins (at 0.25 Hz/bin) adjacent to, but lower than, the frequency of interest. Data points with a S/N ratio estimate of less than 3 were excluded from further analysis.

D. Procedures

Two series of measurements were obtained from the two subjects in this study. The first series consists of ERG measurements at temporal frequencies from 10 to 64 Hz (or 10 to 56 Hz in subject SW, at 100% modulation), each at five retinal illuminances ranging from 650 to 13,000 Td, acquired at either 25% or 100% modulation. The second series consists of measurements of the flicker ERG at 16, 22, 40, and 48 Hz, each with modulation ranging from 1.6% to 100%, in seven logarithmic steps. This data set was obtained at retinal illuminances of 1300, 5000, and 13,000 Td. In a typical daily session we recorded either a frequency series at two illuminances or a modulation series at three illuminances. When running the frequency series, we recorded ERG's at a fixed modulation to all nine frequencies in decreasing order, then increased the illuminance between series. When running the modulation series, we recorded ERG's to a single stimulus frequency at the seven modulations in increasing order, then increased the illuminance between series. We used an ascending modulation series because we found[12] that the response amplitude is minimally af-

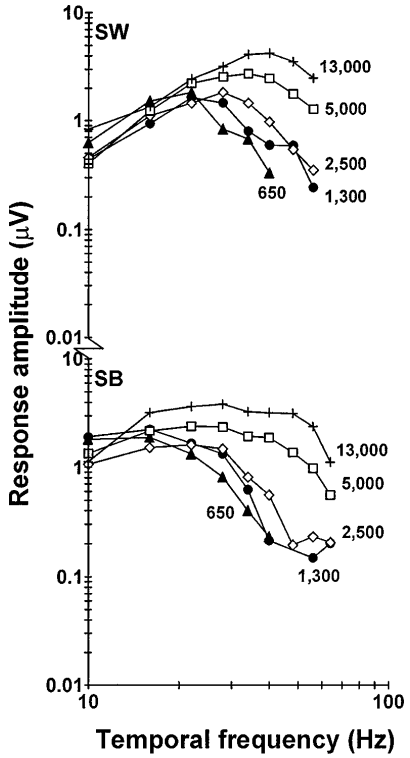

Fig. 1. Response amplitude of the fundamental component of the flicker ERG as a function of temporal frequency, for flickering stimuli at 100% modulation. The number given either to the right of or below each curve indicates the retinal illuminances (in trolands) at which the responses were obtained. Data for the two subjects are shown. The frequencies tested ranged from 10 to 56 Hz for subject SW and 10 to 64 Hz for subject SB. Several high-frequency low-illuminance points were excluded because of the low S/N ratio.

fected by a previous, lower-modulation stimulus but can be strongly affected by a previous, high-modulation stimulus. Between retinal illuminances the subject adapted to the new illuminance while the experimenter set up the new condition, which took at least 30 s. Our illuminance steps were kept to less than 0.7 log unit within any daily session.

E. Data Analysis

We analyzed the responses both in terms of their amplitude at the fundamental frequency and in terms of gain ratio. We defined the gain ratio as the response amplitude produced by a given amplitude of variation in the retinal illuminance. That is, the gain ratio is the response amplitude divided by the mean retinal illuminance, with units of microvolts per troland. We used the term gain ratio to distinguish it from similar but different definitions such as the physiologically defined "gain"[24] and the psychophysically defined "absolute sensitivity"[2,9] or "amplitude threshold."[25]

3. RESULTS

A. Measurements of Response Amplitude and Phase

Figure 1 shows that at 100% modulation the response amplitude was relatively independent of retinal illuminance at low frequencies but clearly increased with increasing retinal illuminance at high frequencies. At higher retinal illuminances the peak sensitivity shifted to higher frequencies. The pattern of results is different for stimuli at 25% modulation (Fig. 2), where at least until 40 Hz the increase of response amplitude with increasing retinal illuminance was less than proportional. The curves relating response amplitude to frequency also show a notch at 40 or 48 Hz at all the retinal illuminances, being deeper at lower retinal illuminances. Results were reproducible. For instance, at 100% modulation the between session variation ranged from 0.07 μV for low-illuminance high-frequency conditions (average response amplitude 0.4 μV) to 1.2 μV for high-illuminance low-frequency conditions (average response amplitude 5.5 μV), with an average of 0.3 μV across all conditions and both subjects (average response amplitude 2.4 μV). This amount of variability is in agreement with the previous results obtained with the same experimental setup.[23] In addition, the differences between runs were correlated and generally represented a change in overall response amplitude rather than a change in the shape of the response functions.

Response amplitude increased systematically with increasing stimulus modulation (Fig. 3). For all the stimulus conditions a power-law relation fitted the data adequately (all $r^2 \geq 0.95$). At 16 Hz the response amplitude was related to the stimulus modulation with an exponent between 0.7 and 0.8 at all retinal illuminances, representing a compressive relation. At 48 Hz the exponent increased with increasing retinal illuminance, showing a compressive power relation at 1300 Td (0.72 for subject SW and 0.79 for subject SB), an approximately linear relation at 5000 Td (slopes of 1.1 and 0.96), and an accelerating power relation at 13,000 Td (slopes of 1.3 and 1.4). The results at 22 and 40 Hz (not shown) are similar to those at 16 and 48 Hz, respectively.

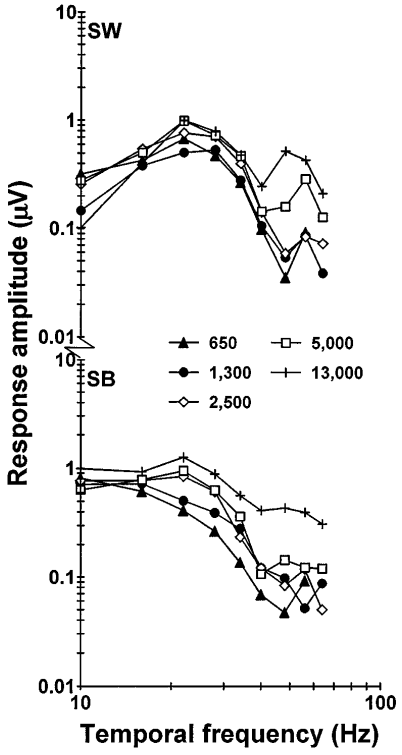

Fig. 2. Same as Fig. 1 but for stimuli at 25% modulation.

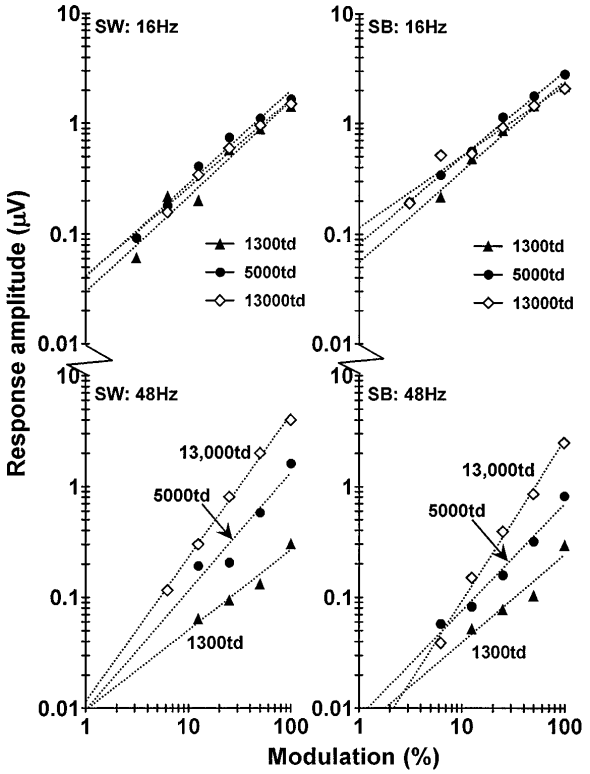

Fig. 3. Response amplitude of the fundamental component of the flicker ERG as a function of modulation depth, for flickering stimuli at 16 and 48 Hz. Data from subjects SW and SB are shown. Within each panel, the three sets of dotted curves represent the best-fitting power law relation to the data at three retinal illuminances. Data points with a S/N ratio of < 3, which occurred occasionally at low modulations, were excluded. All $r \geq 0.95$.

The pattern of results shown by the response phase (Fig. 4) is consistent with the amplitude results. At 16 Hz the phase is relatively independent of modulation, whereas at 48 Hz the phase varies strongly with modulation at both 1300 and 13,000 Td. At an intermediate retinal illuminance of 5000 Td, where the response amplitude was approximately linear with modulation (Fig. 3), the phase variation is also relatively small. For both frequencies there is generally a larger phase shift at higher retinal illuminances.

B. Analysis of the Results by Means of the Gain Ratio

To investigate further how the ERG response is affected by light adaptation we reexamined the data by calculating a gain ratio, which is defined as the ratio of the response amplitude to the mean retinal illuminance. This analysis is analogous to that of Kelly[2] and provides a test of the extent to which the ERG responses follow the predictions of Weber adaptation or linearity. In Figs. 5 and 6 we replotted the data in Figs. 1 and 2 in terms of the gain ratio. At 100% modulation (Fig. 5) the gain ratio shows the classic adaptation pattern, being inversely related to illuminance at low frequencies, consistent with Weber adaptation, and independent of illuminance at high frequencies, consistent with linearity. The linear region, shown by the convergence of the curves at high frequencies, starts at ~40 Hz.

At 25% modulation (Fig. 6) the gain ratio is inversely related to illuminance for frequencies up to 48 Hz, showing adaptation or partial adaptation. At frequencies higher than 48 Hz the gain ratios are less systematic but are clearly not independent of retinal illuminance as predicted by linearity. Indeed, even at high frequencies the gain ratios tend to be larger at lower illuminances, similar to the low-frequency data.

We also analyzed the data in Fig. 3, which show the relation between response amplitude and stimulus modulation, using the gain ratio. At 16 Hz (Fig. 7), the gain ratio is inversely related to illuminance at all modulations. The amount of decrease in the gain ratio with increasing illuminance does not depend on modulation, so the curves are approximately parallel to one another. At 48 Hz the gain ratio is also inversely related to illuminance at low modulations. However, the curves converge at high modulations, where the gain ratio becomes independent of illuminance. The high modulation behavior at this frequency is consistent with linearity. Again the results at 22 and 40 Hz (not shown) are similar to those at 16 and 48 Hz, respectively.

C. Transition from Weber Adaptation to Linearity

To characterize the extent of adaptation at different frequencies, we examined the relation between the gain ratio and retinal illuminance for each frequency, as shown in Fig. 8. For clarity we show results for four representative frequencies only. At 100% modulation (Fig. 8) the gain ratio decreases with increasing illuminance at 16 and 28 Hz (open symbols) but is relatively independent

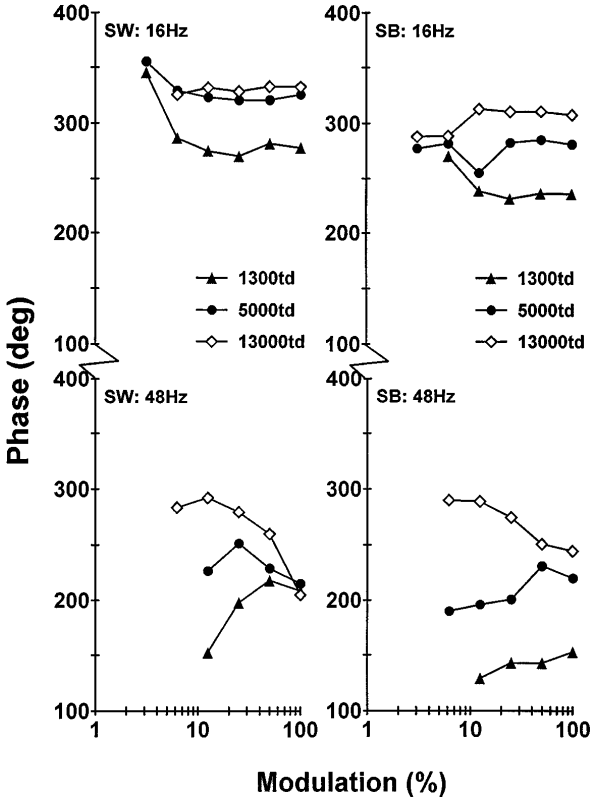

Fig. 4. Phase of the fundamental response component of the flicker ERG as a function of modulation depth, for flickering stimuli at 16 and 48 Hz. These phase measurements were obtained from the same responses shown in Fig. 3. Data from subjects SW and SB are shown. Phase data obtained at the three different retinal illuminances are indexed by the different symbols.

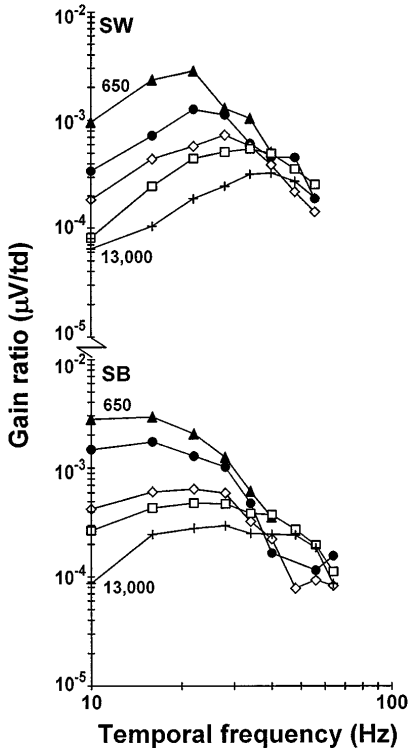

Fig. 5. Gain ratio as a function of temporal frequency, derived from the data shown in Fig. 1. Gain ratio is defined as the ratio of the response amplitude to the mean retinal illuminance at which the response was produced. The five sets of curves within each panel, from top to bottom at the left, represent data at retinal illuminances of 650, 1300, 2500, 5000, and 13,000 Td. Data for subjects SW and SB are shown.

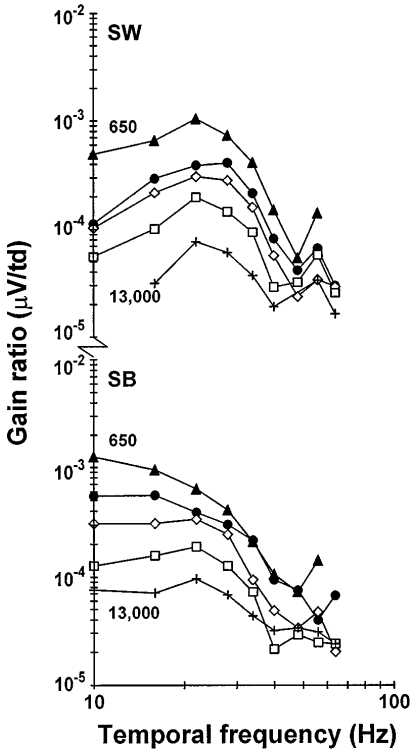

Fig. 6. Same as Fig. 5 but for stimuli at 25% modulation. Gain ratios were derived from the data shown in Fig. 2.

of illuminance at 40 and 56 Hz (filled symbols). Results from other frequencies (not shown) fall between these frequencies. The general trend indicates that the rate of decrease in the gain ratio with increasing illuminance is slower at higher frequencies. At 25% modulation the gain ratio for all four frequencies decreases with increasing illuminance at similar rates. It is not completely independent of illuminance at any frequency.

The dotted curves in Fig. 8 are power-law fits to the data,[26] the slopes of which are used to characterize the extent of adaptation. A power law provides an adequate fit only for the 25% modulation data and for the 100% modulation data at frequencies below 40 Hz ($r \geq 0.86$). At 100% modulation and high temporal frequencies there is not a significant trend in the data that is different from that predicted by the mean. In Fig. 9 we plot the slopes derived from the power-law fits against frequency. At 100% modulation the results show a gradual transition from a slope of −1, corresponding to Weber adaptation, to a slope of 0, corresponding to linearity, with increasing frequency. Linearity occurs at ~48 Hz for subject SW and 40 Hz for subject SB. Note that the slopes derived from the power-law fit to the 100% modulation high-frequency data are variable (and are not significantly different from 0). We include these slopes for completeness of presentation as long as there are at least four data points in the illuminance series. The slope at 48 Hz and 100% modulation for subject SB was omitted, as there were only three data points with a S/N ratio greater than 3 in the series. At 25% modulation, however, the transition either occurs at higher frequencies or never occurs at all. Complete linearity, corresponding to a slope of 0, was not found.

4. DISCUSSION

Our responses obtained at high modulations are consistent with previous findings from both psychophysical[1–3] and electrophysiological[4,5] measurements. However, we found that the responses to low-modulation stimuli show adaptation even at high frequencies. This challenges the notion that gain controls simply fail to keep up with high temporal frequencies. Below, we first consider whether these results can be reconciled with those from our previous study,[12] which were attributed to a single gain control. We then consider a number of other candidate hypotheses.

A. Single Gain Control?

In a previous study[12] we found that the response amplitude of the flicker ERG at 26,000 Td varied with stimulus modulation according to a power law, showing a compressive relation at 16 Hz and an expansive relation at 40 and 48 Hz. The results led us to suggest a modulation-dependent gain control in the flicker ERG. The gain control was hypothesized to be dependent on the low-pass-filtered response. A more elaborate version of such a frequency-dependent gain control model was developed by Graham and Hood.[27] However, the results from the present study suggest that the gain control is also operating at high frequencies. This is inconsistent with any fixed filtering effect.

Another finding in our previous study is that at 26,000 Td one can significantly increase the ERG responses to low-modulation, 40- and 48-Hz stimuli by mixing the 40- or 48-Hz stimulus with another stimulus

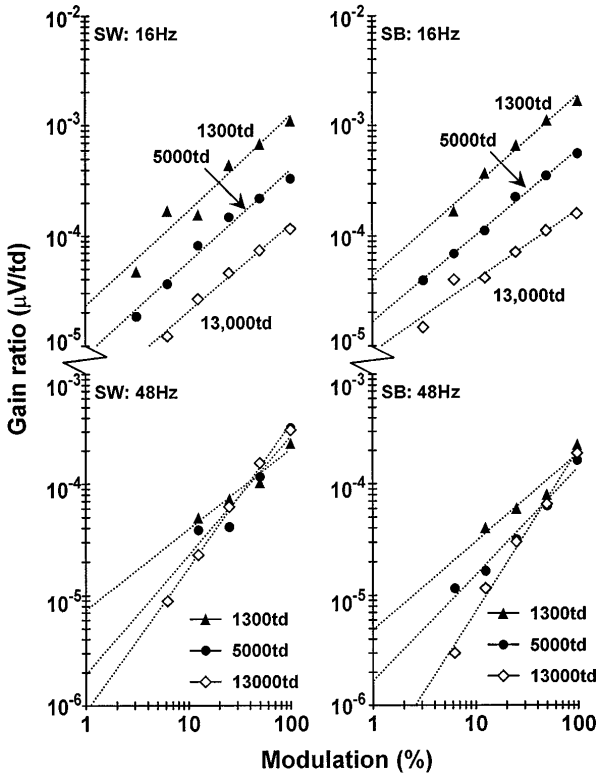

Fig. 7. Gain ratio as a function of modulation depth, for flickering stimuli at 16 and 48 Hz. Gain ratios were derived from the data shown in Fig. 3. Within each panel, the three sets of dotted curves represent the best-fitting power-law relation to the gain ratios at three retinal illuminances. All $r \geq 0.95$.

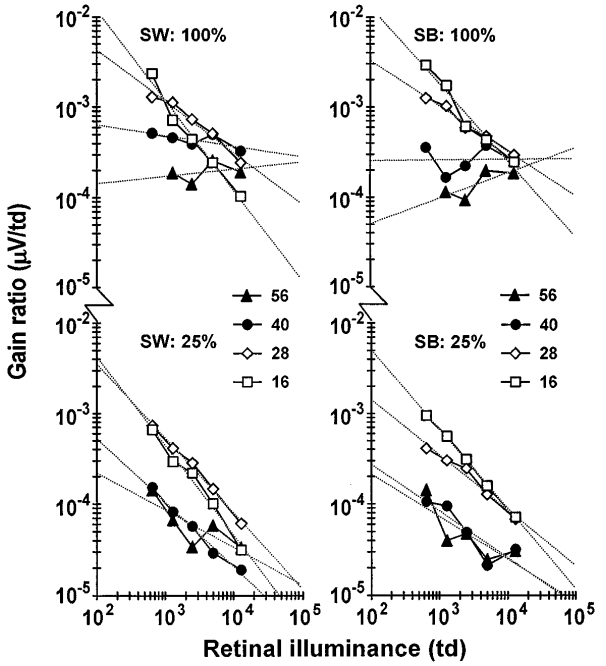

Fig. 8. Gain ratio as a function of retinal illuminance, for flickering stimuli at 100% and 25% modulation. Within each panel, gain ratios are shown for four representative frequencies, 16, 28, 40, and 48 Hz. The dotted curves represent the best-fitting power-law relations to the data at these frequencies. The fits are reasonably good for data at 25% modulation and for data at 100% modulation and low temporal frequencies ($r \geq 0.86$). For data at high temporal frequencies and 100% modulation (e.g., top, solid triangles), we cannot find a power-law relation that predicts the gain ratios significantly better than the mean.

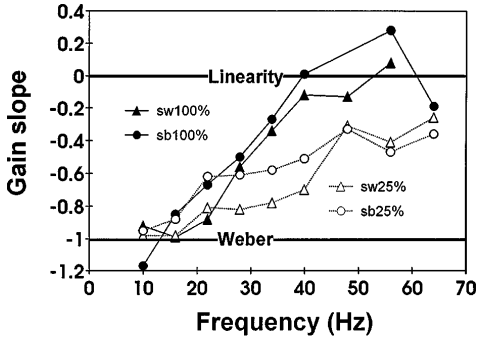

Fig. 9. Slope of the gain ratio versus the illuminance relation as a function of temporal frequency, derived from the power-law fits sampled in Fig. 8. The two thick horizontal lines indicate the slopes of 0 and −1, corresponding to the predictions from linearity and Weber's law. Data from the two subjects were indexed by either triangles or circles, with data at 100% modulation and 25% modulation shown. The slope at 48 Hz and 100% modulation for subject SB was omitted, as there were only three data points with S/N ratio >3 in the series.

of any frequency. This amplitude increase was accompanied by a change in response phase such that the original phase shift with increasing modulation was eliminated or reduced. Similar changes also occurred when the two stimuli were presented successively. These results supported our suggestion of a single-channel modulation-dependent gain control, in which the phase of the control signal is modified by retinal activity. In the present study the amplitude minimum (Figs. 2 and 6) and the phase shift (Fig. 4) found at frequencies close to 48 Hz appear to suggest a transition between two mechanisms. However, before reaching this conclusion we consider whether these results are consistent with the suggestion of an active adaptation process similar to that found in our previous study.[12] If this were the case, then similar frequency interaction effects could be expected. In our control experiments, however, we found that at a retinal illuminance of 1300 Td, the low-modulation, 40- and 48-Hz responses did not increase in the presence of an additional stimulus. This led us to consider explanations other than a single gain control mechanism.

B. Other Possible Mechanism(s)

One explanation of the frequency and modulation dependence of our data is that there are two mechanisms contributing to the ERG response. There are two findings that support this idea: (1) For 25% modulation stimuli the flicker ERG consistently shows a response minimum at 40 or 48 Hz (Figs. 2 and 6). Such a response minimum may reflect a transition between the mechanisms. (2) The phase of the response at 48 Hz (or 40 Hz) also shows a large variation with modulation (Fig. 4), consistent with a transition between mechanisms and cancellation at the transition. In our control experiments we first tested two possible hypotheses for the candidate mechanisms.

First, we tested the hypothesis that rod intrusion produced by scattered light accounts for one of the mechanisms. We varied the surround luminance in five steps over a 70:1 luminance range. ERG responses were recorded to 48 Hz, 25% modulation stimuli at both 650 and 1300 Td. Within the series of five increasing surround luminances the third setting was chosen such that

it approximately matched one of the two retinal illuminances. The results showed no systematic variation in the response amplitude with the surround luminance, rejecting the hypothesis of response cancellation of cone signals by rod signals to scattered lights.

Second, we tested the hypothesis that the response minimum is due to a transition between the long-wavelength-sensitive and medium-wavelength-sensitive cone-mediated responses. If this is so, then the minimum should be shifted or reduced when the color of the flickering stimuli is varied. ERG responses were recorded to red (633 nm), green (543 nm), and yellow (633 and 543 nm) stimuli at 40, 48, and 56 Hz, all at 25% modulation. These conditions were tested at both 2500 and 5000 Td. Although the responses were small and variable, they showed no systematic dependence on the stimulus color. Specifically, when the stimulus color was different from yellow the response amplitude did not increase, as would be expected if we eliminated response cancellation.

We then considered the following two hypothetical temporal frequency-dependent mechanisms, an adaptive mechanism that operates predominantly at low modulations and a linear mechanism that operates predominantly at high modulations. At high frequencies both mechanisms contribute to the response, with their relative contributions depending on modulation. At low frequencies, presumably only the adaptive mechanism contributes to the response. Below we further consider whether any known retinal components are consistent with these hypothetical mechanisms.

First we consider the hypothesis that our response measure, which is represented by the Fourier amplitude of the fundamental component of the flicker ERG, is contributed by both the *a* and the *b* waves of the ERG response. Hood and Birch[28] suggested that the first 10 ms of the cone *a* wave shows no time-course change, very little adaptation, and a small amount of response compression. In contrast, it is generally believed that *b* waves show Weber adaptation.[29,30] Based on these results we assumed that our low-frequency, adaptive response reflects primarily the *b*-wave contribution, whereas our high-frequency, linear response reflects the *a*-wave contribution. If the shapes of the *a*- and the *b*-wave components were adjusted such that they showed a smooth transition with temporal frequency at 100% modulation, then the question is whether it was possible to scale the high-frequency component linearly with modulation in such a way as to generate the marked transition at 25% modulation that we measured. The answer is no. In fact, the modulation dependence of the two components must be sufficiently different to generate the different adaptation patterns at the low and high modulations. If, for example, we assume that the low-frequency component increases with modulation by a power of ~0.7 (consistent with our measurements; see Fig. 3), then the high-frequency component must increase with modulation by a power of ~1.3 to produce a noticeable transition at 25% modulation. (This was confirmed in our preliminary modeling.) If the high-frequency component increases with modulation linearly, then the transition at both low and high modulations will be similarly smooth. Thus our initial hypothesis that there is a linear mechanism that is contributed by *a* waves is contradicted, because an unadapted *a*-wave mechanism, presumably cone photoreceptors, should respond linearly both to mean retinal illuminance and to relative modulation.

Second, we considered the hypothesis that our low-modulation results reflect a transition from central retinal elements to more peripheral elements with increasing temporal frequency. Psychophysically, it has been documented that in the periphery the critical flicker frequency is generally higher[31–34] and flicker sensitivity peaks at higher temporal frequencies[35] than in the fovea. Electrophysiologically, the optimal frequency for the more peripherally distributed M cells is higher than for the P cells,[36] and the temporal sensitivity of the focal ERG is higher in the peripheral retina than in the central retina for temporal frequencies higher than 30 Hz.[37] The latter result[37] has been interpreted as supporting an outer retinal origin of the increased psychophysical temporal sensitivity in the periphery to high temporal frequencies. However, such a center-versus-periphery difference is unlikely to be responsible for our ERG results, as the full-field ERG arises from a spatially summed response, with its amplitude roughly proportional to the stimulated area.[38] Because of this areal summation the central portion of the retina makes a relatively small contribution to the full-field ERG. For example, the central 15° of our 40° field contributes only 14% to the total area. Thus we believe that our 40° responses are generated mostly in the periphery. Although a shift from central retinal elements to peripheral elements with increasing frequency is possible, its contribution to the global ERG response is negligible. Moreover, for a center-to-periphery shift to account for our results, two additional assumptions must be made: (1) the shift must be modulation dependent, which has not been found, and (2) the adaptation properties of the central and the peripheral retina must be sufficiently different, which is in disagreement with the threshold focal ERG results of Seiple and Holopigian,[37] in which the threshold-versus-illuminance curves from both the central and the peripheral retina seem to be consistent with Weber adaptation.

Finally, as both mechanisms involved are presumably nonlinear, we considered the hypothesis that our results are contributed mostly by proximal retina, for instance, by a transition between the M and P cell responses. This possibility has been suggested by Tyler[9] for explaining his psychophysically determined mechanisms. However, we believe that it is unlikely that ganglion cell responses contribute significantly to the flicker ERG, as it is generally believed that the flicker ERG arises from the distal retina (see, e.g., Baker *et al.*[16]), mediated either directly by photoreceptors or by photoreceptors plus bipolar cells (see Section 1). Although a small ganglion cell component can be extracted from the flicker ERG by a nonlinear systems analysis technique,[39] owing to its nonlinear property and local variation this component cannot contribute significantly to a global response. In addition, Tyler[9] has found that the two mechanisms mediated by M and P responses were present only with small-field stimulation, whereas a single mechanism was sufficient for explaining Kelly's 65° data,[2] suggesting that the sustained mechanism can be activated only when there are flickering edges presented near the fovea (by small-field

stimulation). Thus according to this suggestion our 40° stimulus field was too large for the edge-sensitive, sustained mechanism to contribute. Moreover, all the retinal illuminances tested in this study ($\geq$2.8 log Td) belonged to Tyler's high-luminance range. Thus we conclude that our experimental technique and conditions were unsuitable for obtaining both M and P cell responses.

In summary, although there is evidence in our data against two mechanisms, such as the smooth increase of responses with increasing modulation (Fig. 3), our results point to the possibility of a modulation-dependent transition between two mechanisms. Although the physiological substrates for these mechanisms are uncertain, both must depend on modulation nonlinearly. An alternative possibility is that one of the mechanisms, the adaptive one, also adjusts its time course as a result of adaptation and thus combines with the other mechanism with a variable delay. The latter possibility has also been proposed by Hood and Birch.[28]

In conclusion, the results obtained in the present study cannot be fully explained by the single gain control proposed in our previous study. These low-illuminance responses are mediated either by two modulation-dependent nonlinear mechanisms or by one nonlinear mechanism that dynamically adjusts both its sensitivity and its time course with adaptation.

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26. The analysis shown in Fig. 8 is similar to Tyler's luminance analysis (see Ref. 9). However, Tyler fitted the relation between absolute sensitivity (ΔL) and luminance (L) with a template $\Delta L = kL + L_0$, where L_0 represents a low-luminance asymptote. In this study we fitted the data in Fig. 8 by using only the power-law function because our measurements were obtained at retinal illuminances ($\geq$ 2.8 log Td) for which L_0 is unimportant.
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